REMARKS

Claims 50-53, 56-65 are pending in this Application. The Applicant has cancelled claims 51, 54, 55, 59, 60 and 61 without prejudice to his rights to pursue the subject matter of these claims in this or other applications. Applicant has amended claims 52-53 and 56-58. Applicant has added new claims 62-65. No new matter has been entered.

Objections

Claims 60 and 61 are objected to as being in improper form because a multiple dependent claims can not depend from another multiple dependent claim. Applicant has canceled claims 60 and 61 without prejudice, thereby rendering the rejection moot.

35 U.S.C. § 112, 1st Paragraph, Rejection – Written Description

Claims 51, 52, 53 and 59 are rejected under 35 U.S.C. 112, first paragraph, as failing to comply with the written description requirement.

More particularly, the office action indicates that limitation that the blood samples "comprises leukocytes which have not been fractionated into cell types" is new matter.

Applicant respectfully traverses the rejection. However, solely in the interest of expediting prosecution, Applicant has cancelled claim 51, without prejudice, and has amended claims **52** and 53 to remove dependencies to claim 51, without prejudice to the newly cancelled subject matter.

The office action also indicates that claim 59 is rejected for having new matter in its recitation of the phrase "wherein none of said control subjects are subject to systemic steroids or have a disease selected from the group consisting of rheumatoid arthritis, hypertension, obesity, allergies, mild osteoarthritis and severe osteoarthritis". Applicant respectfully traverses the rejection. However, solely in the interest of expediting prosecution, Applicant has cancelled claim 59.

In view of this amendment and remarks, Applicant respectively requests that this rejection be reconsidered and withdrawn.

35 U.S.C. § 112, 2nd Paragraph Rejections, - Indefiniteness

Claim 59 is rejected under 35 U.S.C. § 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which Applicant regards as the invention.

The office action indicates it is not clear to which control subject the instant claim refers to since the independent claim sets forth two different groups of control subjects. Applicant respectfully traverses the rejection. However, solely in the interest of expediting prosecution, and as discussed in the preceding section, Applicant has cancelled claim 59, without prejudice, rendering this rejection moot.

35 U.S.C. § 112, 1st Paragraph Rejections, - Enablement

Claims 50-53 and 56-59 are rejected under 35 U.S.C. 112, first paragraph, as failing to comply with the enablement requirement. Applicant respectfully traverses the rejections.

The instant claims are drawn to a method of classifying netrin 4 (NTN4) gene expression in a human test subject, where the method results in a classification of netrin 4 (NTN4) gene expression in said test subject with that of said subjects having osteoarthritis, or with that of said subjects who are healthy.

The Office action indicates that:

> "The nature of the invention requires the knowledge of a reliable association between NTN4 expression and the ability to classify a particular individual's expression with the expression of subjects having OA or not having OA..."

Applicant asserts that that the specification has discovered an association between NTN4 expression and osteoarthritis (OA). As acknowledged on page 6 of the office action, Example 24 discloses that the identification of the NTN4 gene as being differentially expressed in blood samples from individuals having osteoarthritis when compared with healthy individuals. Specifically, Example 24 discloses:

> "Total mRNA from a drop of peripheral whole blood taken from each patient was isolated using Trizol^{RTM} reagent (GIBCO) and fluorescently labeled probes for each blood sample were generated as described above. Each probe was denatured

> and hybridized to a 15K Chondrogene Microarray Chip (ChondroChip™) as described herein. Identification of genes differentially expressed in blood samples from patients with disease as compared to healthy patients was determined by statistical analysis using the Wilcox Mann Whitney rank sum test (Glantz S A. Primer of Biostatistics., 5th ed. New York, USA: McGraw-Hill Medical Publishing Division, 2002)", paragraph 399, US 2005/0123938, the USPTO publication of the instant specification.
>
> "FIG. 22 shows a diagrammatic representation of gene expression profiles of blood samples from individuals having osteoarthritis as compared with gene expression profiles from normal individuals. Expression profiles were generated using GeneSpring™ software analysis as described herein. Each column represents the hybridization pattern resulting from a single individual. Normal individuals have no known medical conditions and were not taking any known medication. Hybridizations to create said gene expression profiles were done using the ChondroChip™ ..... 300 differentially expressed genes were identified as being differentially expressed with a p value of <0.05 as between the osteoarthritis patients and normal individuals. The identity of the differentially expressed genes is shown in Table 3O", paragraph 400, US 2005/0123938, the USPTO publication of the instant specification.

Table 3O, entitled "Genes Corresponding To Differentially Expressed Genes in Figure 22 – Osteoarthritis", lists netrin 4 (NTN4) as being differentially expressed with a p value = .000163813, and locates it on GeneSpot 1241 of the ChondroChip™. Based on the above expression data and statistical analyses of NTN4 expression in blood samples of osteoarthritis patients and of normal individuals, the specification supports the instantly claimed method of classifying netrin 4 (NTN4) gene expression in a human test subject, with that of subjects having osteoarthritis or with that of subjects who are healthy.

However, Applicant respectfully disagrees with the assertion in the office action that the practice of the invention requires:

> "an understanding of how the presence of osteoarthritis effects the level of NTN4 expression in human blood", page 5 of the office action.

Applicant knows of no enablement requirement that necessitates disclosing mechanisms effecting gene expression with respect to the claimed method of classifying gene expression.

Applicant notes that the instant claims are not drawn to a method of diagnosing osteoarthritis, nor to a method of discriminating between patients with osteoarthritis and rheumatoid arthritis. In contrast, the instant claims are drawn to a simple a method of classifying NTN4 gene expression in a blood sample of a test subject with that of healthy subjects as recited in newly amended claim 58, or with subjects with osteoarthritis, as recited in newly added claim 62. The use of the classification method is not disclosed to be an unequivocal diagnosis, but only one method in a battery of diagnostic assays to contribute to a diagnosis of osteoarthritis.

Accordingly, Applicant also disagrees with the contention in the office action that observing differences in expression of NTN4 between two populations is a highly unpredictable endeavor, which is based on:

> "The specification teaches differential expression of this gene between populations of patients with OA and healthy controls. The specification also teaches that NTN4 is differentially expressed in the blood of patients who have rheumatoid arthritis versus healthy controls patients", page 8 of the office action.

and

> "So even if one carried out the claimed analysis on a test subject, and if one observed a level of expression, it is highly unpredictable how one would begin to know if that level of expression would b e properly classified with patients having osteoarthritis, rheumatoid arthritis, both, one but not the other, something in between or even some other condition or disorder for which the expression profile has not yet been determined", page 9 of the office action.

As discussed above, Applicant emphasizes that the instant claims are not drawn to a method of diagnosing OA, nor a method of detecting OA, nor a method of distinguishing OA from RA. In contrast, the instant claims are drawn to a classification method, which, as suggested in the office action when read in light of the specification, is designed to be used "to provide a tool that is used as part of a diagnostic process". As such, the claimed methods contain no resolution step of diagnosis OA, and leaves open the use of other methods to confirm the diagnosis and/or the extent with which NTN4 is useful as a marker for RA and/or OA.

Regarding the concern of the office action that the claimed methods appear to identify both those with RA, as well as those with OA, Applicant notes that even the

much litigated patented method claims of Metabolite Laboratories, Inc.'s U.S. Patent No. 4,940,658, ('658), include method steps which can be used to indicate a disease or disorder other than the disease/disorder recited. For example, Claim 13 of '658 is drawn to a method for detecting a deficiency of cobalamin or folate in warm-blooded animals by assaying a body fluid for an elevated level of total homocysteine, and is thus used as a method to detect vitamin deficiency. However, it was well known in the medical community before the filing of '658, that the assay for elevated homocysteine levels could signal an increased risk of heart disease. Despite much scrutiny for other reasons, claim 13 of '658 has not been invalidated as a result of other previously known use(s) of its claimed assay to provide a correlation to a second disease or disorder not recited in its claim 13.

Here, the instant claims provide a method to classify NTN4 gene expression either with patients with OA or patients without OA, as part of a diagnostic protocol, presumably playing a role as a preliminary diagnostic tool.

Classification of gene expression with population groups is well established and therefore not an encumbrance to unpredictability. The specification discloses how to classify NTN4 expression:

> "Identification of genes differentially expressed in blood samples from patients with disease as compared to healthy patients or as compared to patients without said disease is determined by statistical analysis of the gene expression profiles from healthy patients or patients without disease compared to patients with disease using the Wilcox Mann Whitney rank sum test", paragraph 0130, US 2005/0123938.

It is well known that the Wilcox-Mann-Whitney two-sample test may be thought of as testing the null hypothesis that the probability of an observation from one population exceeding an observation from the second population is 0.05. Thus, the Wilcox-Mann-Whitney test is a well established and widely used means of identifying differentially expressed genes among populations as instantly disclosed:

> "As would be understood to a person skilled in the art, one can utilize sets of genes which have been identified as statistically significant as described above in order to characterize an unknown sample as having said disease or not having said disease. This is commonly termed "class prediction". Paragraph 0135, US 2005/0123938,

and

> "Methods that can be used for class prediction analysis have been well described and generally involve a training phase using samples with known classification and a testing phase from which the algorithm generalizes from the training data so as to predict classification of unknown samples (see for Example Slonim, D. (2002), Nature Genetics Supp. Vol 32 502-8, Raychaudhuri et al. (2001) Trends Biotechnol 19: 189-193; Khan et al. (2001) Nature Med. 7 673-9; Golub et al. (1999) Science 286: 531-7. Hastie et al. (2000) Genome Biol. 1(2) Research 0003.1-0003.21 all of which are incorporated herein by reference in their entirety)", paragraph 0136, US 2005/0123938.

Thus, Applicant respectfully disagrees with the contention on page 8 of the office action that states that observing differences in expression between two populations is a highly unpredictable endeavor, particularly in the instant method of NTN4 classification.

In maintaining the enablement rejection, the office action contends that "the instant specification has not established that all difference no matter the magnitude nor the direction, relative to any control subjects or even relative to a healthy control subject is related to osteoarthritis". Applicant notes that the only control group in claim 58 as newly amended is healthy controls, and that Example 24 provides full support for the claim as follows:

> "300 differentially expressed genes were identified as being differentially expressed with a p value of <0.05 as between the osteoarthritis patients and normal individuals. The identity of the differentially expressed genes is shown in Table 30", Paragraph 0400, US 2005/0123938.

Given the disclosed data in Example 24 and Table 3O, and well established methods of statistical analysis, Applicant contends that it would not be undue experimentation for one of skill to follow the claimed methods of classification of NTN4 gene expression in a human subject, nor to determine the direction of the change in differential expression. The fact that the instant claims require determination of a statistical similarity or a statistical difference in gene expression levels between the test subject and the control subjects clearly makes it unnecessary to include the direction and magnitude of differential gene expression for enablement. Further, the instant specification discloses that a "post-normalization cutoff of a ratio not equal to 1.0 is used to identify differentially expressed genes", paragraph 0127, US 2005/0123938.

In view of the working examples, data showing differential NTN4 expression between blood samples from healthy subjects and subjects with OA, the disclosed, well established methods of statistical analysis of gene expression, and the disclosed methods, an excerpt of which follows:

> "Identification of genes differentially expressed in blood samples from patients with disease as compared to healthy patients or as compared to patients without said disease is determined by statistical analysis of the gene expression profiles from healthy patients or patients without disease compared to patients with disease using the Wilcox Mann Whitney rank sum test", Paragraph [0130] US 2005/0123938,

Applicant contends that it would not require undue experimentation to practice the claimed method of classifying the expression of NTN4 in a test subject with control subjects

The office action indicates that the scope of the independent claims includes patients who have OA as well as other co-morbidities. Applicant notes that a survey of recently issued claims regarding diagnosis based on differential gene expression rarely, if ever, contains the limitation that the diagnosed patient not be afflicted with an additional disease or disorder. Rarely does a patient have only one condition or disorder.

Nevertheless, in the interest of expediting prosecution of the instant application, Applicant has newly added claim 62, drawn to "*a method of screening a human test subject for being a candidate for having osteoarthritis*", to more clearly set forth the claimed subject matter which Applicant considers to satisfy the enablement requirement.

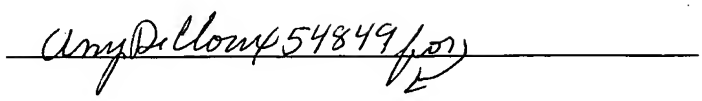
In view of the remarks and claim amendments, Applicant respectfully requests reconsideration and withdrawal of the rejection of the instant claims.

Conclusion

Applicant submits that all claims are allowable as written and respectfully request early favorable action by the Examiner. No new matter is added. If the Examiner believes that a telephone conversation with Applicant's attorney/agent would expedite prosecution of this application, the Examiner is cordially invited to call the undersigned attorney/agent of record.

Respectfully submitted,

Date: June 16, 2008



Name: Kathleen Williams
Registration No.: 34,380
Customer No.: 21874
Edward Angell Palmer & Dodge LLP
P.O. Box 55874
Boston, MA 02205
Tel: 617-239-0100